Environmental Pollution 207 (2015) 280–287

Contents lists available at ScienceDirect

Environmental Pollution

journal homepage: www.elsevier.com/locate/envpol

Effects of environmentally relevant mixtures of major ions on a freshwater mussel

Serena Ciparis [a, *], Andrew Phipps [a], David J. Soucek [b], Carl E. Zipper [c], Jess W. Jones [d]

[a] Department of Fish and Wildlife Conservation, Virginia Tech, Blacksburg, VA 24061, USA
[b] Illinois Natural History Survey, Champaign, IL 61820, USA
[c] Department of Crop and Soil Environmental Sciences, Virginia Tech, Blacksburg, VA 24061, USA
[d] U.S. Fish and Wildlife Service, Department of Fish and Wildlife Conservation, Virginia Tech, Blacksburg, VA 24061, USA

A R T I C L E I N F O

Article history:
Received 18 June 2015
Received in revised form
5 September 2015
Accepted 7 September 2015
Available online xxx

Keywords:
Conductivity
Mining
Appalachian
Bivalve
Unionidae

A B S T R A C T

The Clinch and Powell Rivers (Virginia, USA) support diverse mussel assemblages. Extensive coal mining occurs in both watersheds. In large reaches of both rivers, major ion concentrations are elevated and mussels have been extirpated or are declining. We conducted a laboratory study to assess major ion effects on growth and survival of juvenile *Villosa iris*. Mussels were exposed to pond water and diluted pond water with environmentally relevant major ion mixtures for 55 days. Two treatments were tested to mimic low-flow concentrations of Ca^{2+}, Mg^{2+}, $SO_4{}^{2-}$, $HCO_3{}^{+}$, K^+ and Cl^- in the Clinch and Powell Rivers, total ion concentrations of 419 mg/L and 942 mg/L, respectively. Mussel survival (>90%) and growth in the two treatments showed little variation, and were not significantly different than in diluted pond water (control). Results suggest that major ion chronic toxicity is not the primary cause for mussel declines in the Clinch and Powell Rivers.

1. Introduction

Major ions of geologic origin are fundamental components of freshwater ecosystems. However, anthropogenic activities are causing major ion concentrations to increase in freshwaters worldwide (Williams, 2001). Toxic effects of elevated major ion concentrations to freshwater species have been observed in laboratory studies, and elevated major ions in natural waters have been correlated with aquatic species losses (Canedo-Arguelles et al., 2013; Kefford et al., 2004). Elevated major ion concentrations in freshwaters can induce osmotic stress in freshwater organisms (McCulloch et al., 1993). The toxicity of major ions to freshwater organisms depends upon both the concentration and the ionic composition (Goodfellow et al., 2000; Mount et al., 1997; Soucek and Kennedy, 2005).

Mining is a common cause for freshwater salinization (Canedo-Arguelles et al., 2013; Hancock et al., 2005; Pond et al., 2008; Schreck, 1995), defined as an increase in the total concentration of dissolved inorganic ions in water and estimated by measuring total dissolved solids (TDS) as an evaporative residue (Hem, 1985). Mining operations move unweathered geologic materials into the ambient environment, where exposure to O_2 and H_2O enables accelerated weathering and major ion release (Orndorff et al., 2015). Mining-origin elevated stream salinity and associated alterations of aquatic communities have been documented throughout Appalachian USA (Bernhardt et al., 2012; Cormier et al., 2013; Evans et al., 2014; Griffith et al., 2012; Hitt and Chambers, 2014; Pond et al., 2008; Timpano et al., 2015). The dominant major ions in mining-influenced Appalachian waters are generally $SO_4{}^{2-}$, $HCO_3{}^{-}$, Ca^{2+}, and Mg^{2+}; but K^+, Na^+, and Cl^- can also become elevated (Cormier et al., 2013; Pond et al., 2008; Timpano et al., 2015).

Increasing salinization of Appalachian streams and rivers is of concern because they often support exceptionally high biodiversity. The Clinch and Powell Rivers occur within the Upper Tennessee River system in eastern USA (Fig. 1); both originate in Virginia and flow into northeastern Tennessee. Both river mainstems are established refugia for diverse biotic assemblages, as they remain free-flowing throughout Virginia and their watersheds have low human population densities relative to most other eastern USA areas. The Clinch and Powell Rivers support 49 extant freshwater mussel species, 21 of which are federally endangered (Johnson

* Corresponding author. 106a Cheatham Hall, MC 0321, 310 W. Campus Drive, Virginia Tech, Blacksburg, VA 24061, USA.
E-mail address: sciparis@vt.edu (S. Ciparis).

http://dx.doi.org/10.1016/j.envpol.2015.09.023
0269-7491/

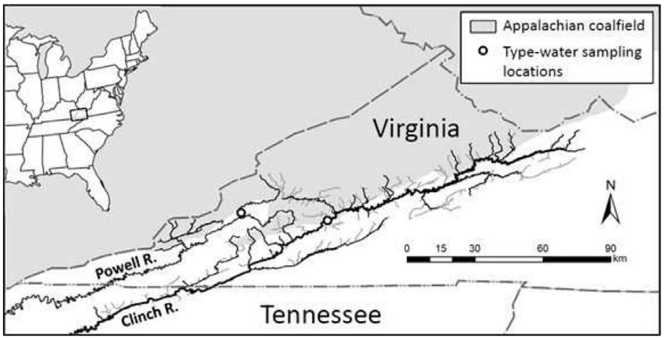

Fig. 1. Locations of the Clinch and Powell Rivers in Virginia relative to the Appalachian coalfield (gray shading). Samples collected from the type-water sampling locations (open circles), Big Stone Gap on the Powell River and Dungannon on the Clinch River, were used to prepare treatments simulating ion concentrations in each river.

et al., 2012; Jones et al., 2014). However, surface coal mining occurs within upper portions of the Powell River watershed and in watersheds of the Clinch River's northwestern tributaries (Fig. 1). Long-term water quality monitoring of the Clinch and Powell Rivers has documented increasing temporal trends of TDS concentrations since the 1960s (Price et al., 2011, 2014). Independent biological monitoring, initiated in 1979, has documented significant declines of freshwater mussel richness and density in sections of Clinch River and throughout the Powell River (Ahlstedt et al., 2005; Johnson et al., 2012; Jones et al., 2014). Recent research has found a spatial association between elevated TDS concentrations and declining freshwater mussel populations in the Clinch River; reaches of the river experiencing the greatest declines in mussel richness and density have elevated concentrations of major ions relative to other reaches of the river where mussel populations are stable (Johnson et al., 2014). In upper segments of the Powell River, TDS concentrations are greater than at any location in the Clinch River (Price et al., 2011) and severe declines of mussel richness and density have been observed in surveyed reaches of the Powell River located downstream of the high TDS reaches (Ahlstedt et al., 2005; Johnson et al., 2012).

This study addresses a fundamental question concerning potential influence by anthropogenic activities on Clinch and Powell River biota: Are major ions acting as toxicants to native mussels? We conducted a laboratory study to assess effects of major ion combinations and concentrations characteristic of the Clinch and Powell Rivers on survival and growth of juveniles of a native mussel species.

2. Materials and methods

2.1. Juvenile mussel production

Juvenile mussels were produced at the Freshwater Mollusk Conservation Center (FMCC), Virginia Tech, Blacksburg, VA following standard propagation procedures (Carey et al., 2013; Zale and Neves, 1982). Gravid female rainbow mussels (*Villosa iris*) were collected from Copper Creek, Scott County, VA on 14 May 2014. Host fish (*Ambloplites rupestris*) were collected from Sinking and Toms Creeks, Montgomery County, VA in early May 2014. Fish were infested with mussel glochidia on 17 May and held in recirculating aquaculture systems at 22 °C. Juveniles excysted from fish hosts 2–3 weeks post-infestation and were cultured in 18 L glass aquariums at 24 °C for approximately 3.5 months prior to study initiation. Juvenile culture methods are detailed in Carey et al. (2013). The water used to hold infested fish and culture juvenile mussels was obtained from a man-made pond at the FMCC which has an on-site well as its source water. Chemical characterization of the pond water was conducted upon initiation of this study (Supplemental Information, Table S-1).

2.2. Test concentrations

The Clinch and Powell treatments were based on data obtained from the Virginia Department of Environmental Quality (DEQ). The highest recorded TDS concentrations (Storet 70300) for impacted sections of the two rivers were 854 mg/L (23 August 2008, at station 6BPOW179.20, Big Stone Gap) for the Powell, and 338 mg/L (5 December 2000, at station 6BCLN237.09, Dungannon) for the Clinch (Fig. 1). The measured TDS values were scaled up to 942 and 419 mg/L (major ion concentration sums), respectively, considering measured alkalinity values as indicators of bicarbonate concentrations, and assuming a 40% volatilization loss of bicarbonate during the evaporative TDS measurement (Howard, 1933). Analyses of other Clinch and Powell River data collected by DEQ, where all major ions and TDS were measured, confirms the ~40% adjustment as appropriate. These data were used to create regressions between TDS and concentrations of individual ions for preparation of treatment waters.

2.3. Treatment preparation

Pond water from the FMCC pond was filtered through a 5 μm polypropylene microfiber filter (Vortex Filter, Filter Specialists, Inc., Michigan City, IN). Undiluted filtered pond water (Pond) and a 50:50 mixture of filtered pond water: deionized water (1/2 Pond) were used as controls and as base waters to prepare treatments. Two different base waters were used to make the two TDS treatments because background ion concentrations in Pond water were too high for synthesis of simulated Clinch River water. The Clinch treatment was 1/2 Pond water with major ions added, and the Powell treatment was Pond water with major ions added. Nominal ion concentrations (Na^+, K^+, Mg^{2+}, Ca^{2+}, Cl^-, SO_4^{2-}, and HCO_3^-) for the Clinch and Powell treatments (Table 1) were based on the regressions of measured TDS concentrations and individual ion concentrations obtained from DEQ data. Recipes for the Clinch and Powell treatments included ion concentrations measured in their respective base (control) waters. Controls and treatments were prepared weekly. Treatments were prepared from base waters using certified American Chemical Society (ACS) reagent grade salts. Potassium chloride (KCl), potassium bicarbonate ($KHCO_3$), sodium bicarbonate ($NaHCO_3$), magnesium sulfate heptahydrate ($MgSO_4$*$7H_20$), calcium chloride dihydrate ($CaCl_2$*$2H_2O$), and sodium sulfate (Na_2SO_4) were purchased from Fisher Chemical (Fair Lawn, NJ). Calcium sulfate ($CaSO_4$) was purchased from Sigma–Aldrich (St. Louis, MO). Ion concentrations in the base waters were verified prior to preparation of treatment waters. A rain event during week 2 of the exposure required adjustment of the recipes to elevate HCO_3^-. Otherwise, ion concentrations in the base waters (and resulting recipes) remained constant. Salts were mixed into 90 L of base water in a 150 L vat with a conical bottom and held for 24 h prior to water exchanges. Each control water (90 L) also was held for 24 h. All waters in the vats were aerated and heated to the target exposure temperature (25 °C).

2.4. Mussel exposure system

Mussels were held in 18 L downweller-bucket systems (Barnhart, 2006); photographs are available in Carey et al. (2013). Each bucket was an experimental unit and held six chambers containing juvenile mussels. Chambers were constructed from